Commonwealth of Virginia
Department of General Services
Division of Consolidated Laboratory Services
Richmond, Virginia

Carbapenem Resistant Organism (CRO) Testing Instructions

• CURRENT DCLS LABORATORY TESTING

Test Type	Method
Identification Confirmation	MALDI-TOF (Bruker Biotyper)
Antimicrobial Susceptibility Testing (AST)*	Broth Microdilution/Sensititre
Phenotypic testing - Carbapenemase Production	Modified Carbapenem Inactivation Method (mCIM)
Resistance Mechanism Identification	Real-Time PCR

*** Submitting laboratories are REQUIRED to provide AST results when submitting suspected CRO isolates to DCLS for testing. These AST results are assessed upon receipt to determine suitability for testing and potential pan-resistance.**

TESTING METHODS

- **mCIM –** Phenotypic testing for carbapenemase production will be performed for all Carbapenem Resistant Enterobacterales (CRE) and Carbapenem Resistant Pseudomonas aeruginosa (CRPA) isolates.
- **AST** – The following drug Minimum Inhibitory Concentration (MIC) values will be determined for all Carbapenem Resistant *Acinetobacter baumannii* (CRAB), as well as mCIM positive CRE and CRPA isolates:
  1. Amikacin
  2. Ampicillin (CRE only)
  3. Ampicillin/sulbactam (CRE/CRAB only)
  4. **Aztreonam* (CRE/CRPA only)**
  5. Cefazolin (CRE only)
  6. **Cefepime***
  7. **Ceftazidime***
  8. Ceftazidime/avibactam (CRE/CRPA only)
  9. Ceftolozane/tazobactam (CRE/CRPA only)
  10. Ciprofloxacin
  11. Ertapenem (CRE only)
  12. Gentamicin (CRE only)
  13. **Imipenem***
  14. Levofloxacin
  15. **Meropenem***
  16. **Piperacillin/tazobactam****
  17. Tobramycin
  18. Tetracycline (CRE/CRAB only)
  19. Trimethoprim/sulfamethoxazole (CRE/CRAB only)

*** Testing required by the CDC's Antibiotic Resistance Laboratory Network (ARLN) for CRO isolates.**
**** Testing required by the CDC's ARLN for *Pseudomonas aeruginosa* and *Acinetobacter baumannii* isolates.**

- **Real-time PCR -** Analyses will be performed on the following resistance marker genes for CRAB isolates and mCIM positive CRE and CRPA isolates:

CRE/CRPA	CRAB
• KPC • NDM • VIM • OXA-48-like • IMP	• KPC • NDM • VIM • OXA-48-like • IMP • OXA-24/40 • OXA-23 • OXA-58

• SPECIMEN TYPES

- Pure suspect CRE isolates on slant or plate media.
    1) Member of the Enterobacterales family
    2) Resistance to at least 1 carbapenem:
        - imipenem, meropenem or doripenem = **MIC of ≥4 µg/mL**
        - ertapenem = **MIC of ≥2 µg/mL**

Note: *Morganella spp.*, *Proteus spp.*, and *Providencia spp.* have intrinsic elevated MICs to imipenem. MIC results for meropenem, doripenem, and/or ertapenem must be used to determine if these organisms should be forwarded to DCLS for testing.

- Pure suspect CRPA isolates on slant or plate media.
    1) Isolate identified as *Pseudomonas aeruginosa*
    2) Resistance to at least 1 carbapenem:
        - imipenem, meropenem or doripenem = **MIC of ≥8 µg/mL**
    3) Mucoid *Pseudomonas aeruginosa* isolates will NOT be tested

- Pure suspect CRAB isolates on slant or plate media.
    1) Isolate identified as *Acinetobacter baumannii* complex
    2) Resistant to at least 1 carbapenem:
        - imipenem, meropenem or doripenem = **MIC of ≥8 µg/mL**

• SPECIMEN TRANSPORT

- Ship isolates at room temperature.
- Submit a completed DCLS Test Request Form with isolate, to include the following:
    1) Complete patient information (including address)
    2) Complete submitter information
    3) Date of specimen collection
    4) Time of specimen collection
    5) Specimen source
    6) Organism suspected
    7) On page 2 of the DCLS test request form check the box for "Carbapenem Resistant Organism" under "Bacteriology ID/ Detection"
    8) Outbreak ID number (if applicable)
- Include copy of laboratory's susceptibility results in specimen package
- **Shipping Address:**

    Division of Consolidated Laboratory Services
    Attn: CRO Team
    600 North 5th Street
    Richmond, Virginia 23219

- **RESULTS REPORTING**
    - Alert value results (**results possibly requiring additional reference laboratory testing**) will be verbally reported within 1 working day of results. An alert notification will be sent to the CDC as well.
    - Non-alert-value, positive results (**results NOT requiring additional reference laboratory testing**) will be verbally reported to the submitting laboratory within 2 working days of results.
    - Negative results will not be verbally reported.
    - A hard copy report of final test results will be provided to the submitter by mail.
        - CRAB isolates will include: identification, AST, and molecular results
        - mCIM positive CRE/CRPA isolates will include: identification, AST, phenotypic, and molecular results
        - mCIM negative CRE/ CRPA isolates will include: identification and phenotypic result only
    - VDH will receive electronic reporting of results.
    - Reference laboratory results for additional characterization testing will be provided to the submitting laboratory and VDH within 2 working days of receipt from the reference laboratory.

- **CONTACT INFORMATION**
    - **For questions regarding testing, specimen collection or shipment, please contact one of the following:**
        - Kristin DiBiase (804-648-4480 x: 339)
        - Dr. LaToya Griffin-Thomas (804-648-4480, ext. 281)
        - Microbial Reference Laboratory (804-648-4480, ext. 287 or 251)
        - Molecular Detection and Characterization Laboratory (804-648-4480, ext. 298)
        - DCLS 24/7 Emergency Mobile Number (804-335-4617)

- **RESOURCES**

Updated information on CRO testing can be found on the DCLS website, under the "Hot Topics" link:
https://dgs.virginia.gov/division-of-consolidated-laboratory-services/updates/hot-topics/

Link to DCLS Test Request Form:
https://dgs.virginia.gov/division-of-consolidated-laboratory-services/resources/submission-forms/

Link to CDC Antimicrobial Resistance Laboratory Network (ARLN) webpage
https://www.cdc.gov/drugresistance/ar-lab-networks/domestic.html